MISCELLANEOUS.

Upon the Mode of Formation of the Egg and the Embryonic Development of the Sacculinæ. By M. E. van Beneden.

In a note inserted in the 'Comptes Rendus' of the month of February last (February 22, 1869), M. Gerbe has given the results of his researches upon the constitution and development of the ovarian egg of the *Sacculinæ*. According to this author, the ovules are formed at their first appearance of two transparent cells closely applied to each other, each provided with a vesicular nucleus and a common membrane (vitelline membrane). One of these cells increases considerably, there are developed in it a large quantity of refractive globules; whilst the other remains small and only acquires a few fine globules; and when the egg is mature, the large cell, in which the elements of the yolk are developed, has attained such a predominance that the other lobe, of which the development has remained in some sort stationary, only represents a small, scarcely perceptible eminence upon one of the poles of the ovule. M. Gerbe regards the large cell as giving origin to the vitellus, and compares it to the yolk of the egg of birds; whilst the little cell, in his opinion, represents the germ or cicatricula. Moreover M. Gerbe thinks he finds in the development of the ovum of the Sacculinæ the explanation of the physiological part performed in the egg by that body which Von Wittich, Von Siebold, and V. Carus have described in the egg of several spiders, by the side of the vesicle of Pürkinje, and which M. Balbiani has observed in certain Myriopods. One of the two cell-nuclei of the primitive bilobed ovule of the Sacculinæ would be the nucleus of the formative cell of the vitellus and the homologue of the vitelline nucleus of the egg of the spiders; the other would be the germinative nucleus or the nucleus of the germ-cell. the homologue of the germinal vesicle of the egg of the spiders and Myriopods.

The observations which I have made upon the development of the ovarian egg of the *Sacculinæ* agree, in certain points, with those of the learned embryogenist of the College of France; but the interpretation which I have given to the facts is essentially different, which is due to the circumstance that, upon some points, I am not able to confirm the investigations of M. Gerbe, and that some important facts have escaped his attention.

The ovules are not, at their first appearance, formed of two cells closely applied to each other; they consist, at first, of a single cell, formed of a perfectly transparent viscous matter (protoplasm) holding in suspension some globules which strongly refract the light, and of a vesicular nucleus, with very delicate outlines, measuring about half the diameter of the cell and provided with a single very refractive nucleolus. The diameter of this cell is about 0·06 millim. Along with these cells are seen others, which present an elongated form and are provided with two nuclei, without, however, manifesting any tendency to the division of their bodies. Others, on the contrary, present at one of their poles a little bud, the size of which increases until it becomes equal to that of the maternal cell; one of the nuclei passes to the interior of the bud, and thenceforward we may recognize two cells, separated from each other by a circular constriction, which deepens gradually; the two daughter cells become individualized, but remain closely connected with each other. The two cells, therefore, are produced by division from a primitive mother cell. I have always found it impossible to distinguish any trace of cell-membrane about these young ovules.

It is indispensable here to make two observations:—first, that these mother cells occur in great quantity in the ovaries immediately after oviposition, as may be seen from the fact that the ovisacs contain eggs which are still at the first commencement of embryonic development; secondly, that the dimensions of the mother cells are the same as those of the little cells which are found in the form of an eminence situated at one of the poles of the mature egg. All the other characters of the mother cells are identical with those which are presented by these polar cells of the mature eggs. In both we see a perfectly transparent protoplasmic body, holding in suspension some rounded, very refractive globules, and we even find some of considerable size, which present no difference in character from those of the vitellus. We distinguish in them a vesicular nucleus, with very delicate outlines, provided with a nucleolus endowed with considerable refractive power.

The mother cells of which I have spoken give origin, by means of division, to two daughter cells. At the commencement of their development these cells are all exactly alike. Soon, however, their bulk increases slightly, and each of them acquires by degrees the dimensions of the mother cell. They each contain some refractive globules; but soon the number of these globules increases greatly in one of the two united cells, and, at the same time, its size begins to exceed that of its congener. From this moment it becomes impossible to distinguish, in the midst of these refractive globules, the nucleus of the enlarged cell. I have never afterwards succeeded in distinguishing in the mature egg the cell-nucleus in the midst of the vitelline mass. The cell increases more and more; it completely fills itself with refractive globules, of which the size increases as quickly as the number. Whilst enlarging, this cell (which we may now call the *egg*, since we recognize distinctly in its contents the characters of a true vitellus) preserves a perfectly regular spheroidal form; only at one of its poles the second cell, which has remained stationary in its development, is attached. When the egg has attained a diameter of from 0·16–0·18 millim. we distinctly recognize in it a cell-membrane, which is developed at the expense of the periphery of the protoplasm of the ovicell, and betrays itself by its dark outline. This membrane (vitelline membrane) is not a common envelope of the enlarged cell (which is the egg in course of development) and of the transparent cell joined to one of its poles; it does not enclose this latter cell, but, on the contrary, its contour stops at the margin of the surface of attachment of the egg and the polar cell. When the egg has arrived at maturity, it presents a regular ellipsoidal form, and at one of its poles the polar cell is always found, retaining its hemispherical form and its original transparency and dimensions. This cell is outside the vitelline membrane, of which we can follow the perfectly regular dark outline between the vitellus and the polar cell on a level with the surface of attachment; the vitelline membrane, however, is slightly depressed, and perhaps it is wanting, at the centre of this surface.

Along with these mature eggs which bear near their poles a transparent cell, are others in which it is not possible to distinguish any polar cell, but which still present, at one part of their surface, a depression corresponding to the old surface of attachment; the extruded eggs never show the least trace of the polar cell, or anything which resembles a cicatricula. Considering this fact in conjunction with that of the existence in the ovary, a little time after oviposition, of isolated cells, which I have called *mother cells*, and which present all the characters of the polar cells of the mature eggs, we see that *the polar cells of the mature eggs are not a constituent part of the egg, comparable to the cicatricula of the egg of birds; these cells separate from the surface of the mature eggs, remain in the interior of the ovary, and increase in number by division to give birth to two daughter cells, which remain attached to each other, and of which one produces in its turn an egg.* The body which M. Gerbe has regarded as representing a vitelline cell, destined to form the nutritive elements of the vitellus, is in reality the entire egg; its nucleus represents the germinal vesicle; and its contents consist of a homogeneous protoplasmic liquid, holding in suspension some refractive globules (nutritive elements of the vitellus).

These observations suffice, it appears to me, to justify the conclusion that I draw from them; but I find in the analogies which the development of the eggs of the *Sacculinæ* present to those of a great number of other Crustacea, and in the development of the embryo of the *Sacculinæ*, the complete demonstration of the conclusion which has just been formulated.

In a great number of parasitic Copepoda (*Caligus*, *Clavella*, *Lernanthropus*, *Congericola*) the ovary presents the form of an oval sac (*germigene*), of which the anterior extremity is prolonged into a tube (*vitellogene*); the latter gradually widens and opens exteriorly, after having formed in the interior of the body a certain number of convolutions. The germigene is filled by a very slender transparent band, twisted and coiled upon itself, which at the entrance of the gland is produced into the tube which represents the vitellogene. This cord is really formed of an immense number of small perfectly transparent protoplasmic cells provided with a very small nucleus. They are flattened, and resemble little disks piled together. In the vitellogene each of these little cells increases in size, and becomes filled with refractive elements, to become an egg, at the same time that their nucleus becomes the germinal vesicle. The eggs retain this flattened discoidal form, and they are accumulated in the vitellogene like coins. In some other Lernæidæ (*Anchorella*, *Lerneopoda*) the division of the ovary into germigene and vitellogene does not exist; but this organ is formed of a ramified tube, of which all the branches are filled with fragments of protoplasmic cords, the characters of which are identical with those of the protoplasmic cords of *Clavella* and *Congericola*. *If the walls of the ovary are torn, a great number of eggs are set at liberty, each of which bears at one of its poles a fragment of protoplasmic cord formed of piled-up discoidal cells.* When the eggs have arrived at maturity they separate from the cord, are ejected, and it is the cell of the protoplasmic cord which was immediately adjacent to the egg that increases, becomes filled with refractive elements, and becomes in its turn an egg. It is impossible not to recognize that these eggs, bearing at one of their poles a fragment of ovarian cord, are really the analogues of the eggs of the *Sacculinæ* provided with a polar cell. The polar cell represents anatomically and physiologically the fragment of the protoplasmic cord of *Anchorella* and *Lerneopoda*, which separates, like it, from the mature egg to furnish new eggs.

In studying the first phases of the embryonic development of the Sacculinæ, *I have ascertained that these animals present at first the complete segmentation of the vitellus.* Now, as I have shown in a previous memoir, the complete segmentation of the vitellus only takes place when the whole mass of the nutritive elements occurs in suspension in the protoplasm of the ovicell, which excludes the idea of a cicatricula. A cicatricula exists when a great part of the nutritive elements is outside the protoplasm of the ovicell, as in birds. In this case these elements do not take part in the division of the ovicell, and the segmentation is partial; it occurs at the expense of the cicatricula exclusively. But in the *Sacculinæ* the whole mass of the vitellus becomes divided into two equal portions, in consequence of the formation, all round the small section of the egg, of a furrow which starts from the periphery and advances gradually towards the centre. Soon afterwards a new furrow appears on the surface of the vitellus, crossing at a right angle that which had first appeared. The mass of the vitellus is thenceforward divided into four portions; they have each the form of a quarter of an ellipsoid which has been divided by two perpendicular planes both passing through the centre. From this moment in each of the four segments a separation takes place between the protoplasmic element and the nutritive elements of the vitellus. The protoplasm of the four segments, carrying with it their nuclei, moves to one of the poles of the egg, which is the extremity of the diameter in which the two planes intersect. We see the four segments become more and more clear at this point, and free themselves completely from the nutritive elements, which are driven to the opposite poles. Then the clear parts, each provided with a nucleus, are separated by a furrow from the darker portion of the segment; they constitute the four first embryonic cells, in the form of little protoplasmic globes, each provided with a nucleus. The four large dark spheres, formed of very refractive elements, no longer represent cells; they will also become fused together, so as to form a single mass of nutritive elements. The embryonic cells, on the contrary, multiply by division, to form a cellular zone of increasing extent, which finally, under the form of a cellular vesicle, will enclose the central mass of nutritive matter. From that time the blastoderm is formed.

It results from this that the large cell, which M. Gerbe has regarded as representing the body producing the vitellus, is really the entire egg,—that the egg of the *Sacculinæ* cannot be compared to the egg of birds, since it is impossible to distinguish in it any parts corresponding to the yolk and the cicatricula,—that the polar cell, which has been considered to represent the germ, is analogous to the protoplasmic cord of the egg of the *Anchorellæ*,—and that this cell separates from the mature egg, and remains in the ovary to become divided there and give origin to new eggs.

It is very evident, also, that no comparison can be established between the vitelline body of the eggs of some spiders, or of certain Myriopods, and the cell-nuclei of the double egg of the *Sacculinæ*. The vitelline body of the egg of the spiders, of which MM. von Wittich, von Siebold, and V. Carus have studied the constitution and the mode of formation, and of which M. Balbiani has proved the existence in the Myriopods, never presents the characters of a vesicle or of a cell-nucleus. This body, far from being general in all the animal series, does not exist in all the Araneida, nor even constantly in the same species of Myriopod, such as *Geophilus simplex*: the signification of this accidental element of the egg remains still to be determined.—*Comptes Rendus*, tome lxix. November 29, 1869, pp. 1146–1151.

Food of Oceanic Animals.

Dr. Wallich complains that I omitted to notice what he had published on the subject. I must confess that I overlooked it.

In his 'North-Atlantic Sea-bed' (p. 131), he says that it may be asked "under what other conditions than exceptional ones can marine animal life be maintained without the previous manifestation of vegetable life, as must be the case if it exists at extreme depths?" and he answers this inquiry by submitting that "in the majority of the marine Protozoa, as, for instance, in the Foraminifera, Polycystina, Acanthometræ, Thalassicollidæ, and Spongidæ, the proof of these organisms being endowed with a power to convert inorganic elements for their own nutrition rests on the undisputed power which they possess of separating carbonate of lime or silica from waters holding these substances in solution." But surely this is not a satisfactory answer to the inquiry. A limpet separates carbonate of lime from sea-water; but it cannot be assumed that this animal (which is well known to be a vegetable-eater) has also the power of converting other inorganic substances for its own nutrition. Foraminifera, as well as Amœbæ, are usually considered animal-eaters, feeding by means of their pseudopodia or expansions of the sarcode. As regards sponges, we find, from Dr. Bowerbank's Monograph (vol. i. p. 122), that, in the greater number, their nutri-